

Tel: (617) 665-7333 support@kactusbio.us kactusbio.com

Pyrophosphatase, Inorganic, GMP-Grade

Catalog #GMP-PYR-YE101

Storage Condition $-20^{\circ}\text{C} \pm 5^{\circ}\text{C}$ for 24 months. Avoid repeated freeze/thaw cycles.

Form Liquid

Source An *E. coli* strain that carries the gene for pyrophosphatase, Inorganic from *Saccharomyces cerevisiae*

Storage Buffer 20 mM Tris-HCl, 100 mM KCl, 0.1 mM EDTA, 1 mM DTT, and 50% Glycerol, pH 8.0

Concentration 0.1U/µL

Unit Definition One unit is defined as the amount of enzyme that will generate 1μ mol of orthophosphate per minute from inorganic pyrophosphate (PPi) in a 10 min standard reaction at 25°C containing 20 mM Tris-HCl, pH 8.0, 2 mM MgCl2, and 2 mM PPi.

Product Contents

Pyrophosphatase, Inorganic (0.1U/μL)

Product Description

Pyrophosphatase, Inorganic catalyzes the hydrolysis of inorganic pyrophosphate to form two orthophosphates. Pyrophosphatase, Inorganic enhances RNA or DNA synthesis by preventing the accumulation of byproduct PPi generated in the reaction.

Applications

- Improving RNA yield in In vitro transcription (IVT)
- Enhancing DNA amplification reaction

Quality Control Statement

This product has been filed with the FDA Drug Master Files and is assigned DMF # 038030. KACTUS manufactures this product according to GMP guidelines and performs stringent quality control testing before release. The production is antibiotic- and animal-free.

Quality Control Release Criteria

Assay	Criteria	
Activity (Enzyme Catalytic Efficiency)	≥ 126U/mL	
Purity (SEC-HPLC)	≥ 95%	
Residual Nickel Salt	≤ 10 ppm	
Residual Heavy Metal	≤ 10 ppm	
Endotoxin	≤ 1EU/mL	
Residual RNase	Negative	
Residual DNase	Negative	

Residual Protease	Negative	
Residual Host Cell Protein	≤ 20ng/mg	
Residual Host Cell DNA	≤ 100pg/mg	
Bioburden	≤ 1CFU/10mL	

Protocol for In Vitro Transcription

1. Prepare the following reaction mixture:

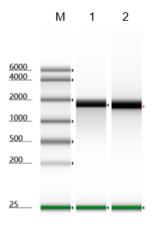
Reagent	Quantity
5X Transcription Buffer-1 (included with T7 RNA Polymerase)	4µL
CTP/GTP/ATP/UTP (100mM each)	2µL each
Murine RNase Inhibitor	1µL
Pyrophosphatase, Inorganic	1µL
T7 RNA Polymerase	2µL
Template DNA	1µg
RNase-free Water	Up to 20µL

- 2. Incubate at 37°C for 1-2 hours.
- 3. After transcription, add 2U <u>DNase I</u> to digest DNA template for 15 minutes at 37°C.
- Inactivate DNase I by phenol/chloroform extraction.

Notes

- Pyrophosphatase, Inorganic works well in any buffer containing Mg²⁺ (1-10 mM)
- The hydrolysis of PPi is proportional to the concentration of the enzyme.

Performance Validation



The addition of Pyrophosphatase, Inorganic to IVT reactions improves mRNA yield. Lane 1: Without Pyrophosphatase, Inorganic; Lane 2: With Pyrophosphatase, Inorganic.